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The MIQE Guidelines' tenth anniversary: The good and bad students

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ABSTRACT

The Polymerase Chain Reaction (PCR) is the most valuable tool that marked the history of molecular biology since the discovery of the DNA structure by Watson and Crick. Its impact on human health over the last 30 years had lead researchers in the field to put strict rules referred to as the Minimum Information for publication of Quantitative real-time PCR Experiments (MIQE) guidelines since 2009. The aim of the current analysis is to shed light on the practice of applying and citing the original MIQE in the published articles over the last decade.

We showed that qPCR is a global technique, but the usage of the MIQE is still lagging in the emerging economies around the globe. We have shown that researchers following the MIQE have better chances of publishing highly cited papers. The MIQE represent the laws for this technique: they enslave us, molecular biologists, into a strict path with financial burdens, but they free us from the "human-errors" a machine would impose on us. As science seeks perfection especially when dealing with human problems, the MIQE, as assessed by the publications' quality over the years, did indeed achieve this step forward towards making the PCR an infallible tool.

1. Introduction

Since the discovery of the DNA structure by Watson and Crick, nothing has been engraved in the field of molecular biology as the Polymerase Chain Reaction (PCR). As most of the landmarks in the history of Sciences, this technique which was first turned down by a prestigious scientific journal was proven to be the most powerful technique in a scientist's toolbox. Few techniques have had the successful story of the PCR and its impact on basic research, and human health. The astonishing evolution from the first generation (PCR) to the "Gold standard" second-generation (quantitative Real-Time PCR, qPCR), until the third and breathtaking generation the digital PCR (dPCR), has beyond any doubts contribute to its transcending usage in every laboratory all over the five continents. The ripeness of these techniques was accomplished by the publication of the Minimum Information for publication of Quantitative real-time PCR Experiments (MIQE) guidelines for the qPCR (Bustin et al., 2009) and the dPCR (Huggett et al., 2013). The main objectives of those guidelines were not to point out the pitfalls in experiments but mostly to help researchers publish high-end reproducible papers. To help with compliance, an application was developed for mobile devices, tablets, and personal computers (Abdel Nour et al., 2014b). One of the major challenges faced by researchers in applying these guidelines was the potential increase in the budget allocated to this technique in their projects. It took only four years down the line to show that respecting the MIQE guidelines in large clinical and pre-clinical trials would have a non-significant increase in the cost of qPCR experiments (Dooms et al., 2013). As a result, the use of the MIQE guidelines shouldn't be an obstacle or a major challenge for researchers.

Recently, the quality of articles reporting the use of qPCR was assessed and published in Nature Methods (Bustin et al., 2013) & PLOS One (Abdel Nour et al., 2014a). In both analyses, papers were scrutinized for their compliance with the MIQE guidelines. The results were deceiving, and the conclusions did suggest that a more rigorous review process should be imposed and followed by both the editors and reviewers. This process was indeed applied by some journals namely the BMC series and Biomolecular detection and quantification (Abdel Nour et al., 2014a). In parallel, Biotech companies started to label the qPCR and/or dPCR products that are compliant with the MIQE-guidelines; among others Bio-Rad laboratories, Roche, Thermo Scientific, Sigma-Aldrich, and PrimerDesign (Abdel Nour et al., 2010).

The aim of the current analysis is to shed light on the practice of applying and citing the original MIQE-guidelines in the published articles. Briefly, the Scopus database was screened for the terms "Real-time PCR" and "qPCR", in all articles that were published between 2009 and 2018. We considered only papers published in journals and we

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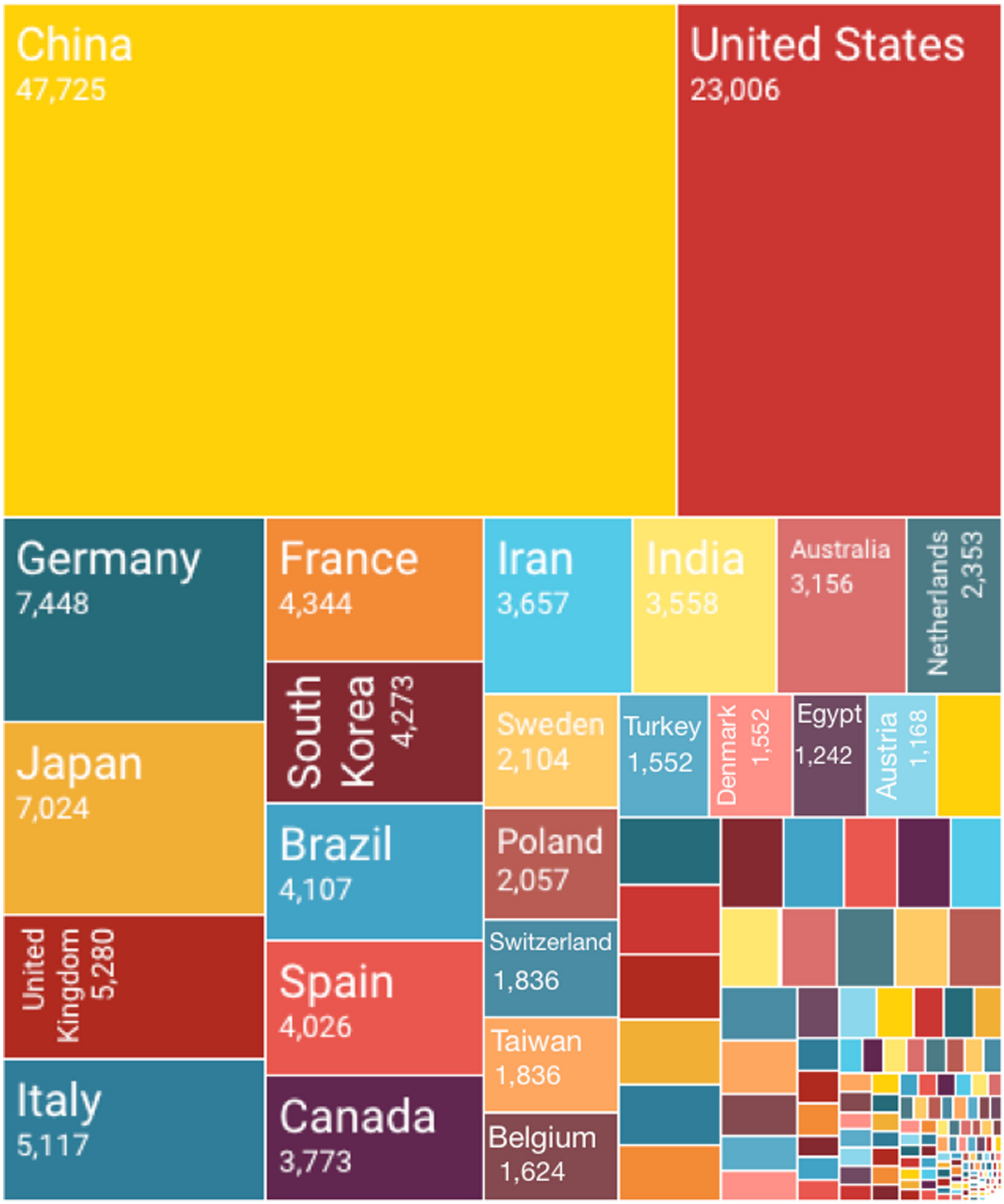

Fig. 1. Number of papers citing "qPCR" between 2009 and 2018 grouped by country.

Table 1
Percentage of papers citing "MIQE" between 2009 and 2018 per affiliated country.

Country	MIQE citing papers
The United States of America	13.6%
China	10.8%
Germany	7.0%
The United Kingdom	5.8%
Brazil	5.2%
Italy	4.6%
Spain	4.5%
France	4.1%
Canada	3.6%
Australia	3.2%

excluded books, books chapters, and conferences. Then we classified those papers by country. The same database was screened for the term "MIQE". The ratio of the "MIQE" over "Real-Time PCR" hits per country was then calculated.

Finally, we agglomerated those articles according to their year of publication and their CiteScore index, a tool developed recently by Elsevier to rank the journals' quality (Roldan-Valadez et al., 2019). We used this metric in order to cross-compare the quality of papers citing "Real-time PCR" or "MIQE" over time and the evolution in the average CiteScore per year.

2. PCR: a global technique

A total of 184 countries and territories have published at least one qPCR paper (Fig. 1). Consequently, qPCR can be considered as a global technique reaching every life sciences laboratory around the globe. Those papers were distributed unevenly per country with China claiming number one with a share of 35.4% of the total number followed by the USA with 17.0% (Fig. 1). The top five western European countries (Germany, United Kingdom, France, Italy, and Spain) could take over the USA with a merged score of 19.4%. Emerging countries (African and the Middle East) are way behind with 3.7%, but it is expected that with the democratization of qPCR, the increase in the

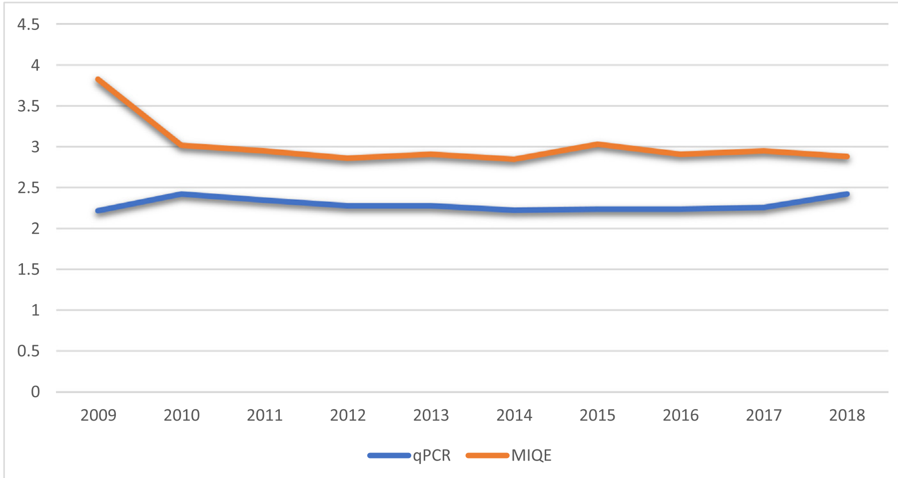

Fig. 2. Mean weighted CiteScore for the "qPCR" and "MIQE" terms between 2009 and 2018.

diversity of products offered by Biotech companies, and the drop in the costs of reagents, the growth rate in the emerging market will be quite spectacular in the coming years. This would be a mirror-image for the efforts made by the Chinese funding agencies in the last decade (2009–2018) which resulted in putting China number in the pole position in the qPCR field with an overwhelming increase of 343.4%.

3. The expansion of the MIQE-guidelines over time

Concomitant with our analysis of "PCR" productivity by country over the last 10 years, we screened the literature for the "MIQE" guidelines which were first introduced in 2009 to enable among others the reproducibility of published experiments. With more than 5977 citations, the MIQE-guidelines are proven to be essential for the qPCR community. However, despite the fact the qPCR users around the world are able to identify and consult the MIQE-guidelines and are convinced of their necessity, few are following them. When it comes to citations of MIQE per country, the distribution is unbalanced. Table 1 shows the top ten countries from where researchers cited the MIQE-guidelines; those countries accounted for 2/3 of the published papers. The USA is first with 13.6%, China is second with 10.8% and the big five Europeans countries totaled an outstanding percentage of 26.1%. The reasons behind this uneven balanced are: i) most of the qPCR experts that published the MIQE guidelines are from Europe, ii) those experts promoted and still are promoting those guidelines in their respective countries and universities/institutions, iii) the unprecedented support from the biotech companies some of which were promoting it either with their MIQE-compliant products or by thousands of training workshops. In contrast, the promotion for the guidelines is still at the embryonic phase in the emerging market. With the exception of a few researchers, the knowledge of those guidelines is still humble in those countries with only 2.7% of the total papers citing the term MIQE.

In our quest to seek the best country that is following the MIQE-guidelines, we calculated the ratio of papers citing the "MIQE" vs "Real-Time PCR". Overall, the worldwide ratio was very low (2.4%), and this result is reflective of the time constraint imposed by the fact that the term "MIQE" was introduced in 2009, whereas the term "qPCR" is there since 1994.

Surprisingly, Georgia has the highest ratio with 100.0%. Obviously, this result is somehow misleading if we take into consideration the total number of published papers on PCR from this country, which turned out to be only one. When considering the top countries, the big five European countries have a cumulative acceptable ratio of 4.0% followed by the USA (2.3%) and China (0.9%).

4. Are the MIQE-guidelines allowing researchers to publish in high CiteScore journals?

The rationale behind having guidelines is to help the scientific community to publish more reliable and reproducible data. In our hunt for an answer for the usefulness of these guidelines in publishing high-quality findings, we compared the mean weighted CiteScore for papers citing "qPCR" with those citing the "MIQE-guidelines" between 2009 and 2018. Fig. 2 shows that throughout the last 10 years, the "MIQE" papers have a higher CiteScore. Thus, researchers following the MIQE guidelines have better chances of publishing highly cited papers.

5. Conclusion

"Law shuttles between freeing us and enslaving us" was one of the eloquent quotes made by the father of PCR, Professor Kary Mullis. The MIQE guidelines represent the law for this technique: they enslave us, molecular biologists, into a strict path with financial burdens, but they free us from the "human-errors" a machine would impose on us. As science seeks perfection especially when dealing with human problems, the MIQE-guidelines, as assessed by the publications' quality over years, did indeed achieve this step forward towards making the PCR an infallible tool.

List of abbreviations

PCR Polymerase Chain Reaction
DNA Deoxyribonucleic Acid
MIQE Minimum Information for publication of Quantitative real-time PCR Experiments
qPCR Quantitative Real-Time PCR
dPCR digital PCR

Contributions

All authors contributed equally to the writing of this article.

Declaration of competing interest

The authors declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of the research reported.

References

Abdel Nour, A.M., Huggett, J., Pfaffl, M.W., 2010. qPCR: current technologies and future applications. European Pharmaceutical Review 15 (2), 17–19.

Abdel Nour, A.M., Azhar, E., Damanhouri, G., Bustin, S.A., 2014a. Five years MIQE guidelines: the case of the Arabian countries. PLoS One 9, e88266.

Abdel Nour, A.M., Azhar, E., Pfaffl, M.W., Damanhouri, G., 2014b. "Stay in touch while on the bench" - how the MIQE applet can increase the quality of your qPCR and dPCR experiments. BMC Genomics 15, 15.

Bustin, S.A., Benes, V., Garson, J.A., Hellemans, J., Huggett, J., Kubista, M., Mueller, R., Nolan, T., Pfaffl, M.W., Shipley, G.L., Vandesompele, J., Wittwer, C.T., 2009. The MIQE guidelines: minimum information for publication of quantitative real-time PCR experiments. Clin. Chem. 55, 611–622.

Bustin, S.A., Benes, V., Garson, J., Hellemans, J., Huggett, J., Kubista, M., Mueller, R., Nolan, T., Pfaffl, M.W., Shipley, G., Wittwer, C.T., Schjerling, P., Day, P.J., Abreu, M., Aguado, B., Beaulieu, J.F., Beckers, A., Bogaert, S., Browne, J.A., Carrasco-Ramiro, F., Ceelen, L., Ciborowski, K., Cornillie, P., Coulon, S., Cuypers, A., De Brouwer, S., De Ceuninck, L., De Craene, J., De Naeyer, H., De Spiegelaere, W., Deckers, K., Dheedene, A., Durinck, K., Ferreira-Teixeira, M., Fieuw, A., Gallup, J.M., Gonzalo-Flores, S., Goossens, K., Heindryckx, F., Herring, E., Hoenicka, H., Icardi, L., Jaggi, R., Javad, F., Karampelias, M., Kibenge, F., Kibenge, M., Kumps, C., Lambertz, I., Lammens, T., Markey, A., Messiaen, P., Mets, E., Morais, S., Mudarra-Rubio, A., Nakiwala, J., Nelis, H., Olsvik, P.A., Perez-Novo, C., Plusquin, M., Remans, T., Rihani, A., Rodrigues-Santos, P., Rondou, P., Sanders, R., Schmidt-Bleek, K., Skovgaard, K., Smeets, K., Tabera, L., Toegel, S., Van Acker, T., Van den Broeck, W., Van der Meulen, J., Van Gele, M., Van Peer, G., Van Poucke, M., Van Roy, N., Vergult, S., Wauman, J., Tshuikina-Wiklander, M., Willems, E., Zaccara, S., Zeka, F., Vandesompele, J., 2013. The need for transparency and good practices in the qPCR literature. Nat. Methods 10, 1063–1067.

Dooms, M., Chango, A., Barbour, E., Pouillart, P., Abdel Nour, A.M., 2013. Improving biological relevancy of transcriptional biomarkers experiments by applying the MIQE guidelines to pre-clinical and clinical trials. Methods 59, 147–153.

Huggett, J.F., Foy, C.A., Benes, V., Emslie, K., Garson, J.A., Haynes, R., Hellemans, J., Kubista, M., Mueller, R.D., Nolan, T., Pfaffl, M.W., Shipley, G.L., Vandesompele, J., Wittwer, C.T., Bustin, S.A., 2013. The digital MIQE guidelines: minimum information for publication of quantitative digital PCR experiments. Clin. Chem. 59, 892–902.

Roldan-Valadez, E., Salazar-Ruiz, S.Y., Ibarra-Contreras, R., Rios, C., 2019. Current concepts on bibliometrics: a brief review about impact factor, Eigenfactor score, CiteScore, SCImago journal rank, source-normalised impact per paper, H-index, and alternative metrics. Ir. J. Med. Sci. 188, 939–951.